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Renal uptake and excretion of homocysteine in rats with acute hyperhomocysteinemia

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Background. Elevated plasma total homocysteine, an independent risk factor for cardiovascular disease, is commonly observed in renal patients. We have previously shown that the kidney is a major site for the removal of plasma homocysteine in the rat. The present investigation was performed to further characterize the capacity of the kidney to handle acute elevations in plasma homocysteine concentrations.

Methods. Acute hyperhomocysteinemic conditions (4- to 7-fold > controls) in rats were produced by either a primed-continuous infusion of L-homocysteine or exposure to 80:20% nitrous oxide:oxygen, which results in the inhibition of methionine synthase.

Results. At physiological homocysteine concentrations, approximately 15% of the arterial plasma homocysteine was removed on passage through the kidney. Renal homocysteine uptake was approximately 85% of the filtered load. The urinary excretion of homocysteine was negligible (<2%). During acute hyperhomocysteinemia produced by the infusion of L-homocysteine, renal homocysteine uptake was increased fourfold and was equivalent to 50% of the infused dose, while urinary excretion remained negligible. Renal homocysteine uptake during nitrous oxide-induced hyperhomocysteinemia increased threefold, with urinary excretion remaining negligible.

Conclusions. These results provide strong evidence that the kidney has a significant capacity for metabolizing acute elevations in plasma homocysteine, and support a very limited role for the re-methylation pathway in renal homocysteine metabolism.

Several recent epidemiological studies have provided strong evidence that an increased total plasma homocysteine concentration is linked to an increased incidence of cardiovascular disease [1, 2]. Homocysteine, a sulfur-containing amino acid, is derived from the intracellular metabolism of the indispensable amino acid methionine. Once formed, homocysteine can be re-methylated back to methionine via the cobalamin-dependent enzyme methionine synthase (EC 2.1.1.13), which requires methyltetrahydrofolate as a co-factor, or via the enzyme betaine:homocysteine methyltransferase (EC 2.1.1.5) [3]. Alternatively, homocysteine can enter the transulfuration pathway and be converted to the amino acid cysteine, through the combined actions of two vitamin B_6-dependent enzymes: cystathionine-β-synthase (EC 4.2.1.22) and cystathionine-γ-lyase (EC 4.4.1.1) [4]. An increase in the plasma concentration of homocysteine likely reflects perturbations in the cellular metabolism of this amino acid, leading to an increased rate of cellular export, a decreased rate of cellular uptake and metabolism, or a combination of these factors.

Several factors are known to lead to increased plasma concentrations of homocysteine, including deficiencies of folate, cobalamin, and vitamin B_6, mutations of enzymes in the metabolic pathway, pharmacological agents [5], and certain disease states. Increased plasma homocysteine concentrations have been documented in human patients with end-stage renal disease [6–8]. These patients are known to be in a higher risk category for atherothrombotic events [9], and the observed hyperhomocysteinemia is likely to play a key role. Poor nutritional status of individuals with renal disease may predispose them to the development of hyperhomocysteinemia [10]. Additionally, the loss of kidney function may signify a role for this tissue in the metabolic clearance of plasma homocysteine. We, in collaboration with other researchers, examined the uptake of homocysteine across the rat kidney and documented significantly higher concentrations of homocysteine in arterial plasma compared to those in the renal vein [11]. The minimal rates of homocysteine excretion observed provided strong evidence that homocysteine was metabolized by renal tubule cells, an observation supported by *in vitro* studies [12, 13]. While the arteriovenous difference for homocysteine was small ($\approx$ 1 μM), it represented 20% of the arterial plasma concentration, positioning the kidney as an important tissue in the metabolic disposal of plasma homocysteine. However, it is not known what reserve capacity the kidney

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possesses to deal with elevated plasma homocysteine concentrations. To address this question, we investigated homocysteine flux across the rat kidney during acute hyperhomocysteinemia, induced by either a primed-continuous infusion of L-homocysteine, or by the exposure of rats to the anesthetic gas nitrous oxide, which inactivates the cobalamin-dependent methionine synthase [14].

METHODS

Animals

Male Sprague-Dawley rats, weighing between 400 and 500 grams, were used throughout this study. The animals were obtained from our institute's breeding colony, and were housed and treated in accordance with the Canadian Council of Animal Care's guidelines [15], as enforced by our institute's Animal Care Committee. Rats were fed a 20% casein-based diet, formulated to the specifications of the AIN-93 semipurified diet [16], designed to meet the nutritional requirements for growth of laboratory rats. The diet and water were available *ad libitum*. Rats were housed at 22°C and exposed to a 12 hour light:12 hour dark cycle, with the dark cycle commencing at 20:00 hours. All experiments were conducted immediately following the termination of the dark phase.

Chemicals

All chemicals (except where noted in the text) were purchased from Sigma Chemical Co. (Oakville, ON, CAN).

Nitrous oxide exposure

For nitrous oxide exposure studies, rats were placed in a cylindrical plexi-glass chamber (15 cm I.D. × 60 cm) through which was passed an 80% nitrous oxide:20% oxygen gas mixture (total flow = 5 liter/min). In previous studies, a mixture of 50% nitrous oxide:50% oxygen gas resulted in a significant reduction in liver methionine synthase activity in rats after 30 minutes [14]. Furthermore, in a recent study [17], rats tolerated exposure to an atmosphere of 80% nitrous oxide:20% oxygen gas for an 18 hour period, indicating that our short term exposure times should be well tolerated. For preliminary time course studies, rats ($N = 3$ per group) were kept in the chamber for either 0 (air breathing controls), 60, 120, or 180 minutes. At the termination of the exposure period, rats were allowed 60 minutes to recover in normal room air. Following the recovery period, rats were anaesthetized with an injection of sodium pentobarbital (60 mg/kg body wt i.p.; Somnitol; MTC Pharmaceuticals, Cambridge, ON, Canada), a mid-line abdominal incision was made and a blood sample taken from the abdominal aorta into a heparinized syringe and immediately placed on ice. Both kidneys were rapidly excised, rinsed with an isotonic saline solution, and frozen in liquid nitrogen until assayed for cystathionine-β-synthase and methionine synthase. Blood samples were centrifuged and the plasma collected and stored at −70°C until analyzed for total homocysteine concentration.

Renal clearance studies

Animals were anesthetized with pentobarbital and surgically prepared for studies of renal clearance, as previously described [18]. Briefly, following cannulation of the trachea, catheters were placed into the left jugular vein (PE-50 tubing; Clay Adams, Parsippany, NJ, USA) and left ureter (PE-10 tubing). A priming dose of 1.75 μCi of [carboxy-^{14}C]inulin (New England Nuclear, Lachine, Quebec, Canada) in 0.8 ml of 5% mannitol-0.45% NaCl was injected into the left saphenous vein. This was followed by a continuous infusion of the same solution, through the jugular catheter, at a rate of 0.037 ml/min, using a Harvard Apparatus model 975 compact infusion pump. Following a 30-minute stabilization period, urine was collected from the ureter catheter into a weighed micro-test tube, for a period of 20 minutes. At the end of the 20-minute collection period, 1 ml blood samples were withdrawn into heparinized syringes from the left renal vein, followed by the abdominal aorta, and the animals were killed by exsanguination. Blood samples were immediately centrifuged at 12,000 *g* for five minutes to remove red blood cells. The plasma was removed and either prepared immediately for the measurement of free homocysteine concentrations or stored at −70°C. Tubes containing the urine were weighed and urinary output was determined as the difference in weights before and after the collection period. Urine samples were then stored at −70°C.

For studies involving L-homocysteine infusion, L-homocysteine was included in the infusion solutions at concentrations calculated to provide 5 μmol/100 g body wt in the prime, and 25 nmol/(min · 100 g body wt) in the continuous infusion. In preliminary experiments, these infusion rates were shown to result in sustained, increased plasma concentration of homocysteine by 30 minutes post-initiation of the infusion (Fig. 1). When homocysteine was included in the infusion solution, an equivalent amount of NaCl (osmotic basis) was removed. L-homocysteine was prepared from L-homocysteine thiolactone, as previously described [19]. For studies involving nitrous oxide exposure, rats were exposed to an atmosphere of 80% nitrous oxide:20% oxygen gas, as described above, for 180 minutes, then subjected to renal clearance studies. Timing of the studies allowed for blood collection at 60 minutes following the termination of nitrous oxide exposure.

Analyses

Plasma total (protein-bound, free-oxidized and free-reduced) and free (oxidized and reduced) and urine total homocysteine concentrations were determined by reverse-phase HPLC and fluorescence detection of ammonium 7-fluorobenzo-2-oxa-1,3,diazole-4-sulphonate thiol adducts, using the method of Vester and Rasmussen [20]. For

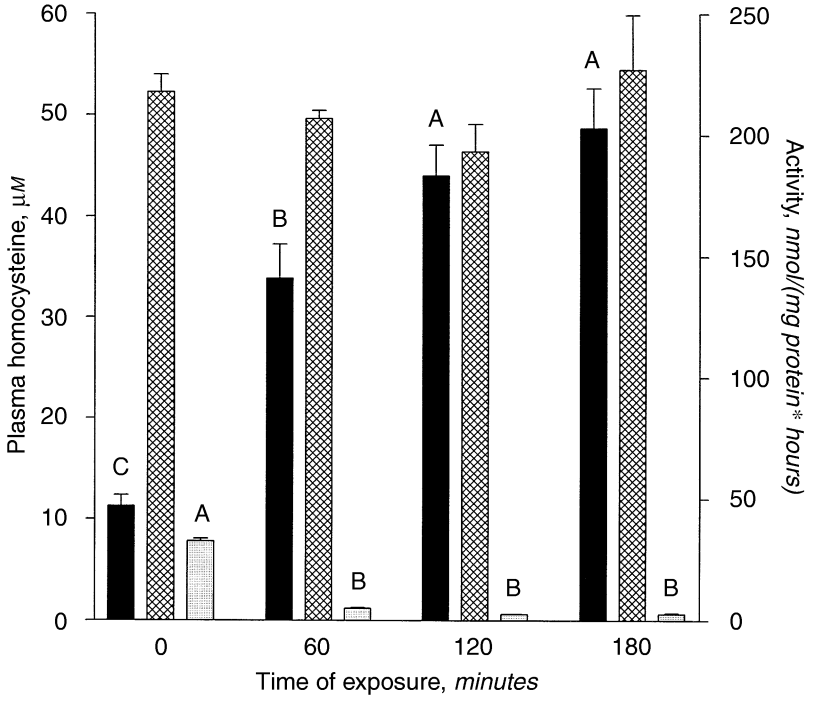

Fig. 1. Effect of exposure time to a 80:20% nitrous oxide:oxygen gas atmosphere on total plasma homocysteine concentrations (μM) and kidney cystathionine β-synthase and methionine synthase activities [nmol/(mg protein · hr) in rats. Data are means plus SEM ($N = 3$ per time point). Different letter superscripts within a bar series denote statistically significant differences ($P < 0.05$) as assessed by ANOVA and Student-Newman-Keul's multiple comparisons procedure. Symbols are: (■) plasma total homocysteine; (▨) kidney cystathione β-synthase; (□) kidney methionine synthase.

determination of ^{14}C-inulin, 25 μl aliquots of plasma and 10 μl aliquots of urine were mixed with 10 ml of a liquid scintillation cocktail (ScintiVerse; Fisher Scientific, Nepean, ON, Canada), and total radioactivity was determined by liquid scintillation counting (Model 1214 Rackbeta LSC; LKB Wallac, Turku, Finland).

For the determination of kidney cystathionine β-synthase and methionine synthase activities, 10% (wt/vol) homogenates were made in 0.05 M potassium phosphate buffer (pH 6.9). The tissue was homogenized with a Polytron (Brinkman Instruments, Toronto, Canada) for 20 s at 50% output and centrifuged at 18,000 × g at 4°C for 30 minutes. Supernatants were collected and assayed for enzyme activities and total protein. Cystathionine-β-synthase activities were measured by the method of Miller et al [21], with cystathionine concentrations measured by reverse-phase HPLC, utilizing pre-column derivatization with o-phthaldehyde and fluorescence detection [22]. Methionine synthase activities were measured by the method of Koblin et al [23]. Protein was measured using the biuret method [24], after solubilization with deoxycholate [25]. Bovine serum albumin was used as a standard.

Calculations

Glomerular filtration rate and renal plasma flow were calculated using the expressions derived by Wolf [26]. Renal uptake of homocysteine was computed as the product of the arteriovenous (A-V) difference for total plasma homocysteine and the renal plasma flow. The filtered load of homocysteine was calculated as the product of the glomerular filtration rate and the arterial plasma concentration of free homocysteine, since it was assumed that homocysteine bound to protein was unavailable for filtration. Homocysteine excretion was calculated as the product of urinary total homocysteine concentration and the urine flow. All values for calculated rates were expressed per 100 grams of rat body wt.

Statistics

Data were analyzed by ANOVA [27]. If the F-test value for the model was significant ($P < 0.05$), treatment means were compared by Student-Newman-Keul's multiple comparisons procedure [28]. All results are presented as the mean (SEM).

RESULTS

Development of acute hyperhomocysteinemia

The primed-continuous infusion of L-homocysteine resulted in a prompt, dramatic rise in its plasma concentration. Thirty minutes following the initiation of the infusion, plasma homocysteine concentrations were approximately sevenfold higher than baseline values [58.06 (3.12) μM versus 7.80 (0.98) μM), and were similar to values obtained after 15 minutes of infusion [62.52 (5.14) μM].

The effect of exposure to 80% nitrous oxide:20% oxygen gas on plasma homocysteine, and the activities of kidney cystathionine-β-synthase and methionine synthase are depicted in Figure 1. Following 60 minutes of exposure (+ 60 min recovery), the concentration of plasma homocysteine was significantly elevated (>3-fold). Furthermore, the specific activity of kidney methionine synthase was reduced to

Table 1. Renal hemodynamic parameters of rats with acute hyperhomocysteinemia resulting from either a primed-continuous infusion of L-homocysteine (+HCY) or 3 hours of exposure to an atmosphere of 80:20% nitrous oxide:oxygen gas (+N_2O)

Parameter	Units	Control ($N = 8$)	+HCY ($N = 8$)	+N_2O ($N = 7$)
Body weight	grams	455 (11)	448 (4)	421 (14)
Urine flow	μl/(min · 100 g)	3.9 (0.4)	3.3 (0.3)	5.1 (0.9)
U:P ratio		232 (33)	252 (19)	213 (33)
Glomerular filtration rate	ml/(min · 100 g)	0.85 (0.11)	0.87 (0.12)	1.02 (0.05)
Renal plasma flow	ml/(min · 100 g)	3.32 (0.49)	3.77 (0.50)	4.13 (0.40)

Rates expressed as unit of volume per minute per 100 g body weight. Values are expressed as means with standard errors presented in parentheses. Results from ANOVA F-test, $P > 0.05$.

Table 2. Renal homocysteine metabolism in rats with acute hyperhomocysteinemia resulting from either a primed-continuous infusion of L-homocysteine (+HCY) or 3 hours of exposure to an atmosphere of 80:20% nitrous oxide:oxygen gas (+N_2O)

Parameter	Units	Control ($N = 8$)	+HCY ($N = 8$)	+N_2O ($N = 7$)
Arterial HCY	μM	7.94^c (0.64)	47.69^a (5.32)	33.42^b (1.13)
A-V Difference	μM	1.14^b (0.32)	5.32^a (1.22)	2.80^b (0.45)
Renal HCY Uptake	nmol/(min · 100 g)	4.12^b (1.65)	18.04^a (3.13)	12.04^a (2.65)
Arterial Plasma Free HCY	μM	5.95^c (0.49)	36.49^a (4.77)	21.89^b (1.00)
Filtered Load of HCY	nmol/(min · 100 g)	5.41^b (1.08)	30.79^a (4.95)	22.21^a (1.45)
HCY Excretion	nmol/(min · 100 g)	0.068^c (0.010)	0.488^a (0.054	0.170^b (0.010)

Rates expressed as nmoles of homocysteine per minute per 100 g body weight. Values are expressed as means with standard errors presented in parentheses. Values within rows with different superscripts are significantly different ($P < 0.05$) by Student-Newman-Keul's multiple comparisons procedure. Renal HCY uptake was calculated as the product of the A-V difference and the renal plasma flow. The filtered load of HCY was calculated as the product of the glomerular filtration rate and the free HCY concentration.

15% of control values. After 120 minutes of exposure (+ 60 min recovery), plasma homocysteine was further elevated (>4-fold vs. controls), while methionine synthase activity was 8% of control values. Plasma homocysteine concentrations and kidney methionine synthase activity in rats exposed for 180 minutes were not significantly different from those values obtained after a 120-minute exposure period. The specific activity of kidney cystathionine-β-synthase in rats exposed to nitrous oxide for any of the time periods was not significantly different from controls.

Renal hemodynamics

Renal hemodynamic parameters are presented in Table 1. There were no significant effects of either L-homocysteine infusion or nitrous oxide exposure on urine flow, glomerular filtration rate, or renal plasma flow. The high U:P ratio, calculated as the ratio of urinary ^{14}C-inulin to arterial plasma ^{14}C-inulin, is indicative of active kidney function, with greater than 99.6% of the glomerular filtrate water being reabsorbed. The values for both glomerular filtration rate and renal plasma flow are consistent with those published previously by our lab [18, 29] and by others [30].

Renal homocysteine metabolism

The arterial plasma homocysteine concentration was significantly elevated by the infusion of L-homocysteine (Table 2). There was a consistent, positive arteriovenous difference for homocysteine, equivalent to approximately 15% of the total arterial plasma homocysteine concentration, in rats not infused with homocysteine (Table 2). In no case was there a zero or negative arteriovenous difference for homocysteine. The infusion of homocysteine resulted in significantly higher arteriovenous difference values for homocysteine, which translated to renal uptake values of homocysteine being significantly elevated by fourfold (Table 2). The arterial plasma free homocysteine concentrations were significantly elevated due to the infusion of homocysteine (Table 2). The mean ± SEM value for the percentage of homocysteine found free in plasma was 75 ± 3%, and was not affected by the infusion of homocysteine (75 ± 5%). The calculated filtered load of homocysteine was also significantly elevated due to homocysteine infusion (Table 2).

The acute hyperhomocysteinemia produced by exposing rats to 80% nitrous oxide:20% oxygen gas for 180 minutes yielded significantly higher renal homocysteine uptake values versus controls (Table 2). Similar to the situation in the L-homocysteine-infused rats, arterial plasma free homocysteine concentrations, and the resulting filtered loads were significantly higher in rats exposed to nitrous oxide compared to controls. The mean ± SEM value for the percentage of homocysteine found free in plasma was 65 ± 2%.

Regression analysis yielded a significant relationship ($P < 0.05$; $r^2 = 0.62$) between arterial plasma total homocysteine concentration and the renal homocysteine uptake of homocysteine (Fig. 2). These data serve to further highlight the ability of the rat kidney to handle acute elevations in plasma homocysteine.

Homocysteine excretion

Both methods employed to generate an acute hyperhomocysteinemia resulted in significantly higher urinary homocysteine excretion rates versus those observed in control rats (Table 2). Despite the higher excretion rates measured in rats with hyperhomocysteinemia, homocysteine excretion accounted for only 1, 2, and 1% of the filtered load of homocysteine for control, L-homocysteine infused, and nitrous oxide-exposed rats, respectively.

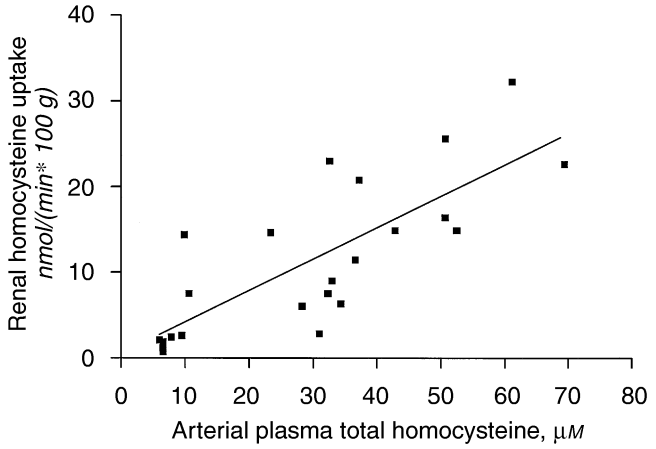

Fig. 2. Regression analysis of arterial plasma total homocysteine concentration versus renal homocysteine uptake. Data represent results from rats from three treatment groups: control, +HCY, $+N_2O$. Regression equation: $y = 0.37x + 0.55$; $P < 0.05$, $r^2 = 0.62$; $N = 23$.

DISCUSSION

The concentration of homocysteine in plasma must necessarily reflect a balance between its rate of appearance in, and disappearance from, the plasma compartment. The results of the present study strengthen our previous observation [11] that, in the rat, the kidney is a major site for the removal of plasma homocysteine. At physiological homocysteine concentrations, approximately 15% of the arterial plasma homocysteine was removed on passage through the kidney. The low rates of urinary homocysteine excretion (<2% of filtered load) strongly indicate that the bulk of the homocysteine taken up by the kidney is metabolized. Furthermore, the renal uptake of homocysteine accounted for approximately 85% of the filtered load, in rats receiving no additional homocysteine. In these animals, renal homocysteine uptake may be limited by the rates of glomerular filtration. In humans, plasma total homocysteine concentrations have been shown to be highly negatively correlated to glomerular filtration rates [31]. The close association between renal uptake and the filtered load of homocysteine observed in the present study may partially explain this clinical finding, however this requires direct experimental confirmation in humans.

During acute hyperhomocysteinemia, induced in the present study by a primed-continuous infusion of L-homocysteine, the renal uptake of homocysteine was increased approximately fourfold over the rates observed at physiological homocysteine concentrations. These results show that the kidney has a substantial reserve capacity to deal with increased plasma homocysteine concentrations. The renal uptake of homocysteine, in the hyperhomocysteinemic rats, when corrected for the physiological rate of homocysteine uptake, was approximately 14 nmol/(min · 100 g body wt), that is, renal uptake (+ HCY) − renal uptake (control) (Table 2). This rate of homocysteine uptake is greater than 50% of the homocysteine infused: 25 nmol/(min · 100 g body wt). These data highlight the importance of renal homocysteine metabolism in relation to the clearance of plasma homocysteine. Despite the substantial uptake of homocysteine by the kidney, renal uptake accounted for 60% of the filtered load during acute hyperhomocysteinemia.

The *in vivo* fate of the homocysteine removed by the kidney remains to be elucidated. We have recently shown that, in incubations of isolated cortical tubules from rat kidneys, greater than 80% of the measured L-homocysteine disappearance was recovered in products of the transulfuration pathway (cystathionine and cysteine) [12]. Methionine appearance accounted for less than 2% of the homocysteine disappearance. Similar results have been reported by Foreman et al [13]. These data, from *in vitro* studies, provide strong evidence that the transulfuration pathway rather than the re-methylation pathway plays the major role in metabolizing the homocysteine removed by the kidney.

In order to explore the importance of the re-methylation pathway to *in vivo* renal homocysteine metabolism, we inactivated the cobalamin-dependent methionine synthase by exposing rats to the anesthetic gas nitrous oxide. Methionine synthase is the sole enzyme responsible for the re-methylation of homocysteine to methionine in rat kidney, due to the lack of betaine:homocysteine methyltransferase activity [32]. The mode of inactivation of methionine synthase by nitrous oxide primarily involves the oxidation of the cobalt atom of the cobalamin group to an inactive state [33], thereby inhibiting the transfer of a methyl group, originating in 5-methyltetrahydrofolate, to the homocysteine molecule. Nitrous oxide may also displace the cobalamin molecule from methionine synthase [34]. We observed a huge reduction in the activity of kidney methionine synthase between 0 and 120 minutes of nitrous oxide exposure. Concurrently, there was a significant increase in the plasma concentration of total homocysteine. Exposure for an additional 60 minutes did not yield further significant changes in either parameter. We also measured the activity of liver methionine synthase and found that, following 60 minutes of exposure to nitrous oxide, the activity was 10% of control values and did not decrease further upon prolonged exposure (data not shown). The observed decrease in methionine synthase activity is consistent with results published by others for *in vivo* studies [12, 15, 35]. Data on the *in vivo* effects of nitrous oxide on plasma homocysteine, especially for short-term exposure studies, are limited [36].

Our results are consistent with the notion that inhibiting methionine synthase activity leads to a rapid increase in the plasma concentration of homocysteine. However, to evaluate whether or not the inhibition of the re-methylation pathway in rat kidney leads to a diminished capacity for homocysteine metabolism in this organ, we measured the uptake of homocysteine across the kidney of rats previously

exposed for 180 minutes to nitrous oxide. There was a significant increase in the renal uptake of homocysteine versus control rats, and this uptake represented 54% of the filtered load, a value consistent with that observed in rats infused with L-homocysteine. These data provide strong evidence that the re-methylation pathway, via the cobalamin-dependent methionine synthase, is not a major factor in the ability of the kidney to metabolize acute elevations in plasma homocysteine. Furthermore, they serve to support our *in vitro* study [12], in that the transulfuration pathway is the primary pathway in the renal metabolism of homocysteine. The activity of the initial enzyme of transulfuration, cystathionine-β-synthase was not affected by exposure of rats to nitrous oxide. Recent studies of whole body homocysteine clearance in folate- or cobalamin-deficient human subjects do not support a major role for the re-methylation pathway in the clearance of plasma homocysteine [37]. In patients with chronic renal failure, whole body homocysteine clearance was 30% of control values, and was not improved with high-dose folic acid supplementation [38]. Thus, in human studies, the kidneys have been suggested to play an important role in clearing plasma homocysteine, and that the re-methylation pathway does not appear to contribute substantially to this clearance. However, direct measures of renal homocysteine metabolism in human subjects are required in order to quantitate the importance of the human kidney for the metabolism of plasma homocysteine.

In the present study, we have calculated the filtered load of homocysteine, assuming ultrafilterability, as the product of the glomerular filtration rate and the plasma-free homocysteine concentration. The percentage of the total homocysteine found in the free form in rat arterial plasma was 65 to 75%. Similar results have been reported previously for rat plasma [39]. These results are in contrast to those reported for human plasma, where the percentage of the total homocysteine found in the free form is $\approx$30% [40]. Since only the free fraction of the total plasma homocysteine should be filtered, the close association we observed, in rats, between the filtered load and the renal uptake of homocysteine, at baseline concentrations, may indicate that a lower fraction of total plasma homocysteine would be available for filtration and subsequent metabolism in the human kidney. Furthermore, homocysteine uptake from the filtrate may partially explain the mode of action of certain pharmacological agents that have been shown to lower plasma total homocysteine concentrations in humans. The thiol agents pencillamine, mercaptoethanesulfonic acid and n-acetylcysteine have been shown to reduce total plasma homocysteine concentrations when administered to humans [41–43]. These agents are thought to out compete homocysteine for thiol binding sites on albumin, thereby resulting in a greater fraction of the total plasma homocysteine being present in the free form [43]. This increase in the free fraction of homocysteine would presumably increase the filtered load and subsequent renal metabolism of homocysteine, without significant increases in the urinary excretion of homocysteine [43], thus leading to lower plasma total homocysteine concentrations.

Concluding remarks

In the present study, we measured renal homocysteine uptake during acute hyperhomocysteinemia, induced by either a primed-continuous infusion of L-homocysteine or the short-term exposure of rats to nitrous oxide. The results obtained provide strong evidence that the kidney has a substantial reserve capacity for metabolizing acute elevations in plasma homocysteine, as is evident from the data in Figure 2. Furthermore, we provide *in vivo* evidence against an essential role for the methionine synthase re-methylation pathway in the renal metabolism of plasma homocysteine during acute hyperhomocysteinemia. These data serve to further characterize the role of the kidney in homocysteine metabolism and position this organ as an important site for the metabolic disposition of plasma homocysteine.

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